Guidance for Analysis and Reporting Under the Revised Total Coliform Rule

Addendum #4

(Revision 1)

to the

Quality Assurance Project Plan (QAPP) for the Texas Commission on Environmental Quality Public Water System Supervision (PWSS) Program Relating to the Safe Drinking Water Act

(Revision 12)

Effective Date

11/4/2017

List of Acronyms

Acronym	Definition
CA	corrective action
CFR	Code of Federal Regulation
COC	chain of custody
E2	Electronic Environmental Drinking Water Reporting System
EPA	Environmental Protection Agency
g	grams
ID	identification
IR	infrared
MF	membrane filter
mg/L	milligrams per liter
mL	milliliters
MRF	Monitoring Reporting Form 10525
MTF	multiple tube fermentation
MUG	4-methylumbelliferyl-β-D-glucuronide
P-A	presence-absence
PDF	portable document format
PWS	public water system
PWSS	Public Water System Supervision
QA	quality assurance
QAPP	quality assurance project plan
QC	quality control
RTCR	Revised Total Coliform Rule
SOP	standard operating procedure
SDWA	Safe Drinking Water Act
SDWIS	Safe Drinking Water Information System
TAC	Texas Administrative Code
TCEQ	Texas Commission on Environmental Quality
TCR	Total Coliform Rule
TNI	The NELAC Institute
UV	ultra violet

Table of Contents

Approval Page – PWSS Program QAPP, Addendum 4

The following TCEQ individuals listed on this page are signatories to this document because they are responsible for the TCEQ's oversight and quality assurance of the work described.

Gary Regner, PWSS Program Quality Assurance (QA) Manager

Texas Commission on Environmental Quality /Office of Water /Water Supply Division

Signature: _____ Date: _____

Gary Chauvin, Manager

Texas Commission on Environmental Quality /Office of Water /Water Supply Division / Drinking Water Standards Section

Signature: _____ Date: _____

Team Leader

Texas Commission on Environmental Quality /Office of Water /Water Supply Division /Public Drinking Water Section /Drinking Water Assessment Team

Signature: _____ Date: _____

Introduction

This document specifies TCEQ requirements related to the analysis and reporting of coliform bacteria in drinking water samples. Laboratories and other entities that generate coliform data—total coliform and *Escherichia coli (E.coli)*—according to these protocols assist the TCEQ in implementing the Safe Drinking Water Act (SDWA). The TCEQ uses the data to make compliance determinations, identify violations, and take assistance actions; thereby protecting public health and ensuring water that is safe to drink.

Requirements in this document are specified for sample collection, chain of custody, analysis, quality control, data validation, and reporting. To submit coliform data to the TCEQ PWSS Program, entities must comply with the criteria and procedures described in this document. The TCEQ reserves the right not to use total coliform and *E. coli* analytical data that do not comply with the specifications defined within this document.

This document reflects changes in requirements as a result of the Revised Total Coliform Rule (RTCR) (40 Code of Federal Regulations (CFR) Part 141.851 and 30 Texas Administrative Code (TAC) §290.109). As in the Total Coliform Rule (TCR), total coliform positive results need to be further analyzed for the presence of fecal indicators. The RTCR uses *E. coli* only as an indicator of fecal contamination, rather than fecal coliform.

Other key modifications in the RTCR which affect laboratories include:

- All provisions related to fecal coliform are removed.
- The 30-hour hold time is more clearly defined. The language in the TCR was vague. The RTCR clearly states that the 30-hour hold time refers to the "time from sample collection to initiation of test media incubation."
- Some changes to the Analytical Methods Table and footnotes were made including but not limited to:
    - The 18th and 19th editions of *Standard Methods for the Examination of Water and Wastewater (Standard Methods)* are no longer approved and applicable references were removed.
    - A clarification to the footnote table indicating that membrane filtration funnels should be sterilized by autoclave, not UV light, prior to beginning the analysis. UV can still be used to sanitize funnels between filtrations. Alternatively, filtration equipment that is pre-sterilized by the manufacturer is also acceptable.
    - Colisure® is defined as a type of media used to perform the reference method SM 9223, rather than a standalone reference method.

The RTCR applies to all public water systems (PWS). Their laboratories must comply with the requirements of the federal RTCR, at <http://www.epa.gov/dwreginfo/revised-total-coliform-rule-and-total-coliform-rule> and the state RTCR, at <https://www.tceq.texas.gov/assets/public/legal/rules/rules/pdflib/290f.pdf>. This document reflects the implementation of the Electronic Environmental Drinking Water Reporting System (E2). The E2 System serves as an electronic filing system, allowing laboratories to manage their own reporting to TCEQ and monitor the status of submitted reports.

Note: *TCEQ rules require that samples be submitted in a manner prescribed by the TCEQ. (TAC §290.46 (b) Electronic data submission is strongly encouraged and may be required by the EPA in the future. The TCEQ will work with laboratories to implement electronic reporting. In the interim, laboratories can still report results manually using the Microbial Reporting Form (MRF) 10525 specified in this document.*

This document is part of the TCEQ PWSS Program QAPP which is reviewed and approved by the

US Environmental Protection Agency (EPA). The requirements in this document are specific to the PWSS Program. They are intended to augment and not supersede requirements contained within the analytical methods/laboratory standard operating procedures (SOP), in The Nelac Institute (TNI) laboratory accreditation standard, and in the *EPA Manual for the Certification of Laboratories Analyzing Drinking Water at* <https://www.epa.gov/dwlabcert/laboratory-certification-manual-drinking-water>.

The current version of this document is located electronically on the TCEQ web page at <https://www.tceq.texas.gov/drinkingwater/microbial/revised-total-coliform-rule>. For questions regarding the implementation of the RTCR, refer to this web page. For specific questions regarding this QAPP Addendum, contact the TCEQ Water Supply Division at (512) 239-4691 and ask for the PWSS Program QA Manager.

Laboratory Requirements/Accreditation

All sample results submitted to the TCEQ under this document must be analyzed by a TCEQ accredited laboratory using methods allowed by the EPA under the SDWA. For questions concerning accreditation, refer to the TCEQ web page located at <https://www.tceq.texas.gov/field/qa/env_lab_accreditation.html>. For specific questions, call 512-239-3754 or email labprgms@tceq.texas.gov.

The TCEQ Laboratory Accreditation Program issued a revision to the drinking water fields of accreditation for microbiology which was effective October 15, 2016. The fields were revised to provide more clarity for the accredited laboratory, to include *E. coli* as an analyte in addition to total coliforms for presence/absence analyses, and to reflect the RTCR. A summary of the changes to the drinking water fields of accreditation for microbiology is located at <http://www.tceq.texas.gov/assets/public/compliance/compliance_support/qa/final_revised_micro_dw_foa.pdf>.

All laboratory accreditation requirements must be adhered to by microbial laboratories, including but not limited to performance testing, data integrity, internal audits, laboratory training, development and maintenance of standard operating procedures, internal data review and management, etc.

Sample Collection

Appropriate sample collection is important to ensure sample results are representative of the water being tested. Although this is primarily a laboratory document, aspects of sample collection are included for the following reasons. (1) It is important for laboratories to understand sample collection protocols so they can provide information to sampling personnel when they obtain their collection bottles and when they submit their samples, (2) sampling collection errors may be cause for sample rejection at the laboratory at the time of receipt, and 3) there are circumstances in which laboratory personnel collect drinking water samples. If the latter case applies, lab personnel must comply with operator certification standards, as discussed below, that apply to the water system type for which they are collecting samples.

Licensing Requirements for Sample Collectors

TCEQ operator-licensing rules stipulate that only a licensed drinking-water operator may collect compliance samples for community and non-transient, non-community systems. There are four classes of certified water operators: A, B, C, and D. Sample collectors must have at least a class D water operator license to collect coliform samples for a community or non-transient non-community public water system. A staff member employed by a transient non-community system may collect compliance samples without a license. See

<http://www.tceq.texas.gov/licensing/licenses/waterlic.html>.

Sample Containers

Sample collection personnel must use laboratory-supplied containers to collect coliform samples. Laboratory-supplied containers are typically 120 milliliters (mL), plastic, and disposable, with a 100 mL graduation mark. Each container must have a durable/waterproof label affixed to the container or a standardized tag attached. The label must have spaces to record the:

- unique sample identification (ID) number,
- sample location,
- date and time of sample collection, and
- sample collector's initials.

Note: *Merely recording the unique sample ID number on a sample collection bottle is not sufficient, even if the ID number can be matched with information on the MRF or the chain of custody (COC) by the laboratory.*

Each container provided by the laboratory must be sterile and contain sodium thiosulfate in either powder, pill, or liquid form to neutralize 5 milligrams per liter (mg/L) of residual chlorine. If sample containers are sterilized in the laboratory, then one sample bottle per batch must be tested for sterility. If any growth occurs during a sterility check, the batch must be re-sterilized. If sample containers are purchased as pre-sterilized, then one bottle per lot purchased must be tested for sterility or at a set percentage such as 1 to 4%.

Laboratories are also required to check and record the effectiveness of the dechlorinating agent. If water having residual chlorine (measured as free, combined, or total chlorine) is to be analyzed, sufficient sodium thiosulfate must be added to the sample bottle before sterilization to neutralize any residual chlorine in the water sample.

Note: *If a disinfectant residual is detected in a sample when it is received at the laboratory, the laboratory must reject it for excessive disinfectant being present.*

Laboratories are also required to check the accuracy of the container's 100 mL mark and auto-fluorescence properties (if used for fluorescence testing), once per lot. The results of all quality control (QC) checks must be documented and maintained by the lab.

A certificate of analysis provided by a vendor may be used to address the container testing requirements of this section. The certificate must include lot specific language; a specification sheet for a product type is not sufficient. If certificates are not supplied with the containers from the vendor, or laboratories prepare their own containers, then the bottle testing requirements described in this subsection apply.

Sample Collection Procedures

For information on proper sample collection procedures and precautions, refer to the TCEQ Regulatory Guidance (RG) 421–*Coliform Sampling for Public Water Systems* at <https://www.tceq.texas.gov/publications/rg/rg-421.html>, and the analytical methods addressed in Section 9060 of *Standard Methods.* At a minimum, sampling personnel **must** measure and record the chlorine residual in the field at each sample site. Samples **must** also be collected consistently with the sample siting plan, as applicable, leaving ample air space in the bottle (approximately 2.5 cm) to facilitate mixing at the laboratory prior to taking a chlorine residual and analyzing the samples. Additional details regarding sample collection procedures are specified in RG 421 and the analytical methods.

Sample Submittal Documentation

If a laboratory has not yet transitioned to the E2 reporting system (which is strongly encouraged) and still reports sample results manually to the TCEQ, then sampling personnel

must submit a MRF 10525 (Exhibit 1) to the laboratories with their samples. Instructions for completing the MRF are included with the form in Exhibit 1. The most current version of this form may be accessed electronically at <https://www.tceq.texas.gov/assets/public/permitting/watersupply/pdw/tcr/microbial_monitoring_form%20RTCR.pdf>. Laboratories may customize the MRF to add their name/logo, contact information, and laboratory ID number in the upper right part of the form. Otherwise, the TCEQ reserves the right to not accept samples/results from modified forms.

***Note**: The Laboratory ID number required on the top, right-hand corner of the form is a laboratory specific, ten-digit ID number associated with the Safe Drinking Water Act Information System (SDWIS). It is the same as the laboratory accreditation number, minus the last 4 digits.*

Laboratories may use the MRF as their COC to avoid the use of multiple forms. If the MRF is used as a COC, then the custody transfer information must be completed on the MRF at the time of laboratory receipt. Alternatively, laboratories can use COC forms that fit their operations as long as they follow applicable rules and regulations regarding COCs. Refer to the EPA *Manual for the Certification of Laboratories Analyzing Drinking Water 5th Ed*, *Appendix 1.*

***Note:** If laboratories use the MRF to report data to the TCEQ and also utilize a separate COC, they should submit both documents at the time of monthly reporting as described in the section-Reporting to the TCEQ.*

If laboratories are currently reporting results using the E2 reporting system, then they may use reporting forms and/or COC forms of their own choice as long as they also follow applicable rules regarding COCs. If sample collectors use a submittal form(s) other than the MRF, the laboratories must ensure the alternate form accommodates mandatory fields as indicated in bold text in Exhibit 1 *How to Complete the Microbial Reporting Form (MRF 10525).*

Laboratory Sample Receipt

The laboratory's role in sample receipt is extremely important. Conformance to the requirements documented in this section will help the TCEQ avoid the need to reject sample results; thus, helping PWSs avoid monitoring and reporting violations.

Custody Transfer

When sampling personnel/couriers deliver samples to the laboratory, custody must be relinquished to a sample custodian or designee.

***Note:** If appropriate personnel are not present to receive the samples, they should be locked in a designated area of the laboratory to prevent tampering. The person delivering the samples should make a log entry identifying the samples that were delivered, the date and time of delivery, and where and how the samples were delivered and secured. Laboratory personnel may then receive custody by noting in a logbook, the absence of evidence of tampering, unlocking the secured area, and signing the custody form.*

The sample custodian (or designee) inspects the sample(s) and sample documentation at the time of receipt for any issues necessitating sample rejection. The sample custodian (or designee) measures and records sample temperature and confirms the absence of a chlorine residual in each sample.

***Note:** The absence of a chlorine residual may be confirmed at the time of analysis as specified in the related section below.*

After the sample custodian inspects and approves the sample and sample submittal documentation, both parties (sampling personnel or courier), and the laboratory custodian or designee will **sign** and date the MRF and/or the COC with the time it was delivered.

Specific requirements related to documentation, sample receipt, confirmation of requirements, and sample rejection are detailed below. As noted in these sections, violations of the requirements or conditions are cause for rejecting samples, except as explained in the next sections.

Insufficient Documentation

It is extremely important for the laboratory to check the sample documentation very carefully at the time of receipt because both incorrect and insufficient documentation may result in the rejection of results and/or monitoring or reporting violations for the public water system.

Note: *The laboratory can use some discretion in assisting the sampling personnel/courier with "fixing" errors in the documentation at the time of receipt in order to avoid the unnecessary recollection of samples. For example, if the laboratory courier (or designee) determines that the type of sample is not checked on the MRF, then he/she can inform the sample collector/courier who can check the appropriate sample type while still on the premises. Similarly, if a ground water well ID number is not recorded correctly (i.e., an address is provided instead), the laboratory may ask the sample collector/courier to correct the entry.*

The laboratory custodian (or designee), cannot correct or complete the form himself/herself. It is the responsibility of PWS personnel to fill out the form, sign, and date it. Under no circumstances can the chlorine residual measured in the field be recorded after the sample is delivered to the laboratory. As specified below, the chlorine residual **must** be measured and recorded in the field. Also, under no circumstances can the laboratory modify the form after it has been received, signed, and dated by the laboratory.

Sample Temperature

To measure sample temperature at receipt, the laboratories may use an infrared (IR) sensor, a bottle blank, a cooler thermometer, or another technique to obtain a temperature measurement. Both the recorded temperature and the corrected thermometer temperature should be recorded on the MRF, as applicable. ***Note:*** *Only a place for a representative sample temperature is provided on the MRF; a temperature for each sample is not required.* Sample containers must never be opened at sample receipt to measure the temperature of an actual sample.

Note: *The preferred method for sample transportation is to hold samples in coolers at <10°C during transit to the laboratory. There is not a requirement for thermal preservation or a temperature criteria which applies to these samples. The laboratory should, however, consider the condition of transported samples and question their validity where temperatures are elevated, such that they might affect microbial concentrations in the sample (if any are present).*

Sample Type

The sample type must be documented correctly on the MRF when the samples are received at the laboratory. If a sample type is not checked (or checked incorrectly) at the time of receipt, then the labs must reject the applicable sample (unless the documentation can be corrected by sampling personnel/courier while still on site) and request a replacement. Sample types for compliance include distribution, ground water well, and repeat samples. Non-compliance sample types include construction and special purpose samples. All sample types may also be replacement samples, if they have previously been rejected.

No Chlorine Residual (On Form)

The laboratory must confirm that a chlorine residual was measured and documented in the field. In order to help the PWS avoid violations, the laboratories **must** reject samples without a documented chlorine residual, as measured in the field, and **not report** results to the TCEQ. This is not an item that can be recorded or documented in the laboratory.

Chlorine Present

The absence of a chlorine residual must be confirmed and recorded by the laboratory at the time of receipt (or at the time of analysis, depending on laboratory operations). This requires pouring off a very small aliquot of well-mixed/shaken sample leaving at least 100 mL of the sample remaining for coliform analysis. If a chlorine residual is detected, the sample **must** be rejected by the laboratory*. **Note:** The absence of a total chlorine residual can be confirmed using test strips such as those marketed by LaMotte.*

Sample Holding Time

For the analysis of total coliforms and *E. coli* in drinking water, the time from sample collection to initiation of test media incubation may not exceed 30 hours. All samples received in the laboratory should be analyzed on the day of receipt. If the laboratory receives the sample late in the day, the laboratory custodian or designee must evaluate the collection time to determine if samples can be run the next day. A sample may be refrigerated overnight as long as the sample is placed in the incubator within 30 hours of sample collection. ***Note:** It is the laboratory's responsibility to manage sample delivery to ensure samples are delivered in time to allow compliance with the holding time requirement. If this does not happen, then the laboratory **must** reject the sample(s).*

Invalid Sampling Point/Invalid Sampling Protocol

It is the responsibility of the PWS to ensure samples are collected at valid sampling points using valid protocols. However, if a laboratory identifies an error related to an invalid sampling point or protocol during sample receipt, it can assist the PWS avoid monitoring and reporting violations if it identifies the problem, rejects the sample, and requests a replacement within 24 hours.

Ground Water Well Identification (ID) Numbers

Ground water well ID numbers always begin with the letter G, followed by the 7-digit PWS ID, then the letters, "A", "B", "C", etc. to indicate which well it was. Ground water well ID numbers are often reported incorrectly to the TCEQ on the MRF. Instead of recording the correct ID number, the sampling personnel will incorrectly record an address or just "Well A", "Well B" etc. Laboratories should ensure that ground water well ID numbers are recorded correctly.

Excessive Sample Volume

When collecting a sample, sampling personnel are required to leave ample air space in the bottle (e.g., at least 2.5 cm) to facilitate mixing by shaking at the laboratory prior to measuring the chlorine residual and running the analysis. If a sample is filled to capacity, the laboratory can reject the sample outright. This is especially appropriate when it is a recurring situation with a specific sample collector. Alternatively, if a sample bottle is too full at receipt to allow for proper mixing, the laboratory must not pour off and discard a portion of the sample. Rather, the laboratory must pour the entire sample into a larger sterile container, mix properly, and proceed with confirming the absence of a chlorine residual and running the analysis.

Sample Rejection

There are TCEQ rejection codes corresponding to the sample receipt issues identified above as specified in the Table of Rejection Codes in the Section - *TCEQ Reporting*. In addition to the reason identified above, there are additional causes for rejecting samples which are self – explanatory, like a sample leaked or broke in transit. If a sample needs to be rejected, the laboratory will notify the PWS of the sample rejection immediately (if still on the premises), and no later than the same business day (or the next business day, if after hours); and request a replacement within 24 hours of notification. ***Note:** If the lab rejects a sample for a reason w/o a specific rejection code, the laboratory can use the general code "LR" for "lab rejected" and document the actual reason on the MRF. In the case of electronic reporting, the reason can be documented in the sample comment field.*

Laboratory Equipment and Supplies

The laboratory must have the equipment and supplies needed to perform the methods for which they are accredited. Supplies and equipment (including pH meters, analytical balances, incubators, refrigerators, autoclaves, water baths, temperature monitoring devices, etc.) should be maintained and calibrated according to the TNI Standard and the analytical methods. In addition, the following criteria apply as specified in the *EPA Manual for the Certification of Laboratories Analyzing Drinking Water.* In cases of conflict, the most stringent criteria apply.

- Glass, dial, or electronic thermometers must be graduated in at least 0.5 degree increments or less, and be calibrated quarterly.
- Incubation units must have an internal temperature monitoring device and maintain the temperature specified by the method.
- Membrane filter units must be stainless steel, glass, porcelain, or autoclavable plastic, not scratched or corroded, and must not leak. See Table 1 footnote #4.
- Membrane filters must be approved by the manufacturer for total coliform analysis.
- Pipettes delivering volumes of 10 mL or less must be accurate to within a 2.5% tolerance.
- Graduated cylinders must be accurate to within a 2.5% tolerance.

Sample Analysis

Allowable Methods

All coliform samples must be analyzed by a TCEQ accredited laboratory using EPA-allowable methods under the SDWA. (See Table 1). These methods may change over time and the Code of Federal Regulations (CFR) is the definitive source for allowable methods. Refer to 40 CFR Part 141.852 at <https://www.ecfr.gov/cgi-bin/text-idx?tpl=/ecfrbrowse/Title40/40cfr141_main_02.tpl> which includes the alternative test procedures. ***Note:*** *Only the analytical methods, for which the TCEQ currently accredits, are included in Table 1.*

The RTCR requires that total coliform positive results be further analyzed for the presence of fecal indicators. The RTCR uses *E. coli* only as an indicator of fecal contamination, rather than fecal coliform. Determination of density is not required. ***Note:*** *With the revision of the TCR, the 18th and 19th editions of Standard Methods are no longer allowed. See Note 1 at the bottom of Table 1.*

Table 1. Allowable Methods

Organism	Methodology Category	Method [1]	Citation [1]
Total Coliform	Lactose Fermentation Methods	Standard Total Coliform Fermentation Technique	*Standard Methods* 9221 B.1, B.2 (20st ed., 21st ed., 22nd ed.) [2, 3] *Standard Methods* Online 9221 B.1, B.2–99 [2, 3] *Standard Methods* Online 9221 B.1, B.2–06 [2, 3]
Total Coliform	Lactose Fermentation Methods	Presence-Absence (P–A) Coliform Test	*Standard Methods* 9221 D.1, D.2 (20th ed., 21st ed.) [2, 7] Standard Methods Online 9221 D.1, D.2–99 [2, 7]
Total Coliform	Membrane Filtration Methods	Standard Total Coliform Membrane Filter Procedure	*Standard Methods* 9222 B, C (20th ed., 21st ed.) [2, 4] *Standard Methods* Online 9222 B–97 [2, 4], 9222 C–97 [2, 4]
Total Coliform	Membrane Filtration Method	m-ColiBlue24® Test [2, 4]	
Total Coliform	Membrane Filtration Method	Chromocult [2, 4]	
Total Coliform	Enzyme Substrate Method	Colilert	*Standard Methods* 9223 B (20th ed., 21st ed., 22nd ed.) [2, 5] *Standard Methods* Online 9223 B–97 [2, 5]

Table 1. Allowable Methods

Organism	Methodology Category	Method [1]	Citation [1]
			Standard Methods Online 9223 B–04
		Colisure®	*Standard Methods* 9223 B (20th ed., 21st ed., 22nd ed.) [2, 5, 6] *Standard Methods* Online 9223 B–97 [2, 5, 6] *Standard Methods* Online 9223 B–04
		Colilert -18 ®	*Standard Methods* 9223 B (20th ed., 21st ed., 22nd ed.) [2, 5, 6] *Standard Methods* Online 9223 B–04
		E*Colite® Test [2] Readycult® Test [2] modified Colitag® Test [2]	
Escherichia coli	*Escherichia coli* Procedure (following Lactose Fermentation Method)	*EC*–MUG medium	*Standard Methods* 9221 F.1 (20st ed., 21st ed.) [2] and 22nd ed) *Standard Methods* Online 9221 F.1-06
Escherichia coli	*Escherichia coli* Partition Method	*EC* broth with MUG (*EC*–MUG)	*Standard Methods* 9222 G.1c(2) (20th ed., 21st ed.) [2, 8]
		NA-MUG Medium	*Standard Methods* 9222 G.1*c* (20th, 21st ed.)
Escherichia coli	Membrane Filtration Method	Membrane Filtration using MI medium	EPA Method 1604 [2]
Escherichia coli	Membrane Filtration Method	m-ColiBlue24® Test [2, 4]	
Escherichia coli	Membrane Filtration Method	Chromocult [2, 4]	
Escherichia coli	Enzyme Substrate Methods	Colilert®	*Standard Methods* 9223 B (20th ed., 21st ed., 22nd ed.) [2, 5 ,6] *Standard Methods* Online 9223 B–97 [2, 5, 6] *Standard Methods* Online 9223-B-04
		Colisure®	*Standard Methods* 9223 B (20th ed., 21st ed., 22nd ed.) [2, 5 ,6] *Standard Methods* Online 9223 B–97 [2, 5, 6] *Standard Methods* Online 9223-B-04
		Colilert -18 ®	*Standard Methods* 9223 B (20th ed., 21st ed., 22nd ed.) [2, 5, 6] *Standard Methods* Online 9223 B–04
		E*Colite® Test [2] Readycult® Test [2] modified Colitag® Test [2]	

Table 1: Notes

1. The procedures must be carried out in accordance with the documents listed in 40 CFR §141.852(c). For *Standard Methods*, the 20th, 21st, or 22nd editions may be used. For the Standard Methods Online, the year in which each method was approved by the Standard Methods Committee is designated by the last two digits following the hyphen in the method number. The methods listed are the only online versions that may be used. For vendor methods, the date of the method listed in 40 CFR §141.852(c) is the date/version of the approved method. The methods listed are the only versions that may be used for compliance with the RTCR. Laboratories should be careful to use only the approved versions of the methods, as product package inserts may not be the same as the approved versions of the methods.

2. Incorporated by reference. See 40 CFR §141.852(c).

3. Lactose broth, as commercially available, may be used in lieu of lauryl tryptose broth, if the PWS conducts at least 25 parallel tests between lactose broth and lauryl tryptose broth using the water normally tested, and if the findings from this comparison demonstrate that the false-positive rate and false-negative rate for total coliforms, using lactose broth, is less than 10 percent.

4. All filtration series must begin with membrane filtration equipment that has been sterilized by autoclaving.

Exposure of filtration equipment to ultraviolet (UV) light is not adequate to ensure sterilization. Subsequent to the initial autoclaving, exposure of the filtration equipment to UV light may be used to sanitize the funnels between filtrations within a filtration series. Alternatively, membrane filtration equipment that is pre-sterilized by the manufacturer (i.e., disposable funnel units) may be used.

5. Multiple-tube and multi-well enumerative formats for this method are approved for use in presence-absence determination under the RTCR.

6. Colisure® results may be read after an incubation time of 24 hours.

7. A multiple tube enumerative format, as described in *Standard Methods* 9221, is approved for this method for use in presence-absence determination under the RTCR.

8. The following changes must be made to the *EC* broth with MUG (*EC*–MUG) formulation: Potassium dihydrogen phosphate, KH_2PO_4, must be grams (g), and 4-methylumbelliferyl-Beta-D-glucuronide must be 0.05 g.

Sample Volume

The sample volume analyzed for total coliforms in drinking water must be 100 mL regardless of method used. To ensure accuracy and consistency, it is important that the laboratory obtain precise measurement of the volume of sample to be analyzed.

Sample Confirmation

A total coliform-positive result is based on the confirmed phase if the Multiple Tube Fermentation (MTF) Technique or Presence-Absence (P-A) Coliform Test; or the verified test for the Membrane Filter (MF) Technique if M-Endo medium or LES Endo agar is used. No requirement exists to confirm a total coliform-positive result using Colilert, Colisure, MI agar, E*Colite, m-ColiBlue24, Chromocult, Readycult/Fluorocult, Coliscan, or Colitag test. Also, no requirement exists to confirm a positive *E. coli* test. In those rare cases where a presumptive total coliform-positive culture does not confirm/verify as such, but is found to be *E. coli*-positive, the sample is considered total coliform-positive *E. coli*-positive.

Rejecting Samples at the Time of Analysis

The laboratory may encounter issues with samples at the time of analysis that do not allow it to begin or complete an analysis. These occurrences are frequently referred to as "unsuitable for analysis." Possible issues include, but are not limited to: cloudy or turbid samples, lab accidents such as spilled samples, or exceeding hold time. The laboratory must notify the PWS on the same day it detects the issue and rejects the sample, (or the next business day, if after hours) so the PWS can collect another sample within 24 hours of notification. The laboratory must also report "rejections" to the TCEQ monthly as described in the section—*TCEQ Reporting*.

Rejecting Invalid Sample Results

If there are problems with the analysis that do not allow the lab to come to a conclusive result, then the sample results should be deemed invalid and rejected. The reasons for invalidating a total coliform sample result (unless total coliforms are detected) include but are not limited to the following:

- Production of a turbid culture in the absence of gas production using an analytical method where gas formation is examined (e.g., Multiple Tube Technique).
- Production of a turbid culture in the absence of an acid reaction in the P-A Coliform Test.
- Exhibiting confluent growth or produces colonies too numerous to count with an analytical method using a membrane filter (e.g., Membrane Filter Technique).

***Note**: Another possible reason for invalidating total coliform sample results is when there is a turbid culture after incubation using Standard Methods 9223, but no color change. This has been reported as an issue by some laboratories. In these circumstances, the inhibitors in the media may be overwhelmed by heterotrophic bacteria and the target organisms not allowed to grow. These situations should, but are not required to be reported to the TCEQ, before reporting a*

negative sample result.

If a laboratory rejects a sample result or a laboratory error occurs, the laboratory must notify the PWS on the same day it rejects invalid sample results (or the next business day, if after hours) so the PWS can collect another sample within 24 hours of notification. The laboratory must also report these occurrences to the TCEQ monthly as described in the section—*TCEQ Reporting*.

Electronic Result Reporting

The E2 is a web-based information system that allows drinking water laboratories to electronically submit their data to the TCEQ. E2 users must have an authorized user account, be granted an association with their laboratory, and fill out a participation agreement.

Currently, the E2 system allows laboratories to submit the compliance data from the MR, 10525. E2 allows the laboratory to report compliance data to the TCEQ utilizing two options:

- Online data entry option
- Data upload option

Users of E2 should refer to the guide–*Electronic Environmental Drinking Water Reporting (E2-DWR) System Laboratory User Guide*, Version 2, September 17, 2014, <https://www.tceq.texas.gov/drinkingwater/e2-reporting-system>. Quick User Guides for both options are also included on the TCEQ website.

Contact Information

For questions or concerns regarding E2, the E2 staff can be reached at: <ESubData@tceq.texas.gov>

Minimum System Requirements

Laboratories must be able to access the TCEQ E2 website through the Internet. Typically, such access is available either through a dedicated connection (i.e. local area network) or a modem connection to an Internet Service Provider.

To ensure that all of the features of the E2 system are available, laboratories must use Microsoft Internet Explorer web browser (version 7.0 or higher) or Firefox (version 10.0 or higher). The performance of the E2 system will vary based on the computer internet connection speed, CPU, Operating System, and available memory. The minimum system configuration recommendation is as follows:

- Broadband Internet Connection or higher
- Pentium II processor or higher
- VGA or higher resolution monitor (at least 800 x 600 resolution)
- Microsoft Windows XP or higher
- 256 MB of RAM or higher
- Portable Document Format (PDF) reader for viewing PDF files
- Printer for printing submission in report format and/or copy of record
- Email account

Manual Reporting using the MRF 10525

Electronic reporting is highly encouraged, although manual reporting is still allowed. The method for reporting manual (or hard copy) results to the TCEQ requires the current MRF 10525. ***Note:*** *If manual results are not reported utilizing the current MRF 10525, the samples will be rejected which may cause the PWS to receive a monitoring and/or reporting violation. MRFs must be reported to the TCEQ by the 10th day of the month following sample analysis by mailing them*

to:

Texas Commission on Environmental Quality
Attn: Revised Total Coliform Rule Program
MC 155
PO Box 13087
Austin, TX 78711-3087

Reporting Positive Results

The laboratory must report positive sample results *(*total coliform and/or *E. coli)* to the TCEQ on the same day they are detected using the TCEQ Microbial Monitoring Positive Result Report Form (Positive Result Report Form) in Exhibit 2 of this document. The TCEQ uses the Positive Result Report Form to manually enter positive result data into the Safe Drinking Water Information System (SDWIS) database where compliance violations are determined. This process also generates notification documents that inform the PWS of the incurred violation(s). The process occurs daily and is in direct response to the reported positive results from laboratories.

In addition to reporting positive results to the TCEQ, the laboratories must also report positive results to the PWS on the day they were detected. When providing to the TCEQ, the laboratory must include a copy of the analytical results provided to the PWS with the Positive Result Report Form. The TCEQ uses the analytical results to confirm whether or not the laboratory has informed the PWS of the positive results and verify SDWIS data entries such as PWS ID, PWS Name, Lab ID, sample ID, sampler initials, collection point, collection date and time, chlorine residual level, sample type, type(s) of indicator organisms present.

Note: *In the past, the TCEQ requested that the MRF be reported with the Positive Result Report Form. This is now only required when the laboratories are using the MRF for reporting analytical results. If the PWS is not using MRF to report analytical results, then the laboratory should include a copy of whatever analytical test report is used with the Positive Result Report Form.*

The laboratory must fax the completed TCEQ Positive Result Report Form and the analytical results to the TCEQ at 1-800-239-0237. Alternatively, the form and supporting documentation may be scanned and emailed to the TCEQ at RTCRPOS@tceq.texas.gov.

Reporting Rejected Samples or Results

If the laboratory rejects a sample or result, these occurrences must be reported in all cases, to the TCEQ in the monthly E2 data report or manually on the MRF. This is required so that the replacement sample results can be tied to the original samples and the sample intent is documented. This will ensure the replacement sample will remain the same sample type as the original sample. In other words, if the rejected sample is a routine sample, the replacement will also be a routine sample.

Table 2 lists the Rejection Codes and the reasons for using each code. These codes are used for both electronic and manual reporting.

Table 2. Rejection Codes

CODE	DESCRIPTION
BR	Broken in transit
CL	Chlorine present (in sample)
EH	Exceeded hold time
EV	Excessive volume

FZ	Frozen sample
HB	Heavy bacterial growth
ST	heavy silt or turbidity present
IN	Insufficient sample information
BP	invalid sampling point
IP	Invalid sampling protocol
LA	Lab accident
LR	Lab rejected
LT	Leaked in transit
NC	No chlorine residual (on form)
VO	Volume insufficient

***Note**: In addition to the required TCEQ reporting described in this subsection, the laboratory must notify the PWS immediately, if possible, and no later than the same day (or next business day, if after hours) when the laboratory rejects a sample, so the PWS can collect another sample within 24 hours of notification.*

Samples may be rejected by the laboratory at the time of receipt as described in the Section - *Laboratory Sample Receipt*. Samples or results may be rejected later in the analytical process. This is discussed in the Section—*Sample Analysis*.

Analytical Records

Laboratories are required to maintain easily accessible records related to this QAPP for five years. A change in ownership, merger, or closure of a laboratory does not invalidate this requirement. The client PWS should be notified before disposing of records less than 5 years old so it may request copies if needed. This includes all raw data, calculations, and quality control data. Electronic data must be backed up. If the laboratory changes its computer hardware or software, it should make provisions for transferring old data to the new system so that it remains retrievable.

Corrective Actions (CAs)

Any person involved with work described in this document must initiate a CA if there is deviation from required protocols specified in it and/or referenced documents. The procedure for a CA following the identification of a deviation begins with an investigation to determine the root cause(s). The laboratory must select and implement the CAs that will eliminate the problem and prevent recurrence. Any CAs identified must be appropriate in degree to the magnitude and risk of the deviation. Laboratory QA Officers (or designees) are responsible for assuring that CAs are documented, reported, implemented, and tracked appropriately.

Deviations that require CA include, but are not limited to the following:

- Equipment failure
- Excursions from quality control limits
- Samples lost due to laboratory accidents
- Failure to meet acceptance limits when analyzing EPA Proficiency Test samples
- Holding time exceedances

Most CAs can be accomplished at the point of origin using an established procedure through some combination of the following: repair or replacement of faulty equipment; re-analysis of samples and standards; checking reagents for proper strength; etc. CA procedures/response actions are specified in laboratory SOPs that include required documentation, solutions, and follow-up.

Unique deviations/problems that cannot be corrected by the procedures listed above will require CAs to be defined when the need arises.

If laboratory deviations involve the following list, the laboratory QA Officer must notify the TCEQ by phone or e-mail within 48 hours, draft a CA report, and submit it to the PWSS Program QA Manager within 14 days of the incident detection.

- Calls into question the integrity of sample analysis results which have been previously reported to the TCEQ
- Results in non-conformance with state or federal regulations
- Was associated with the intentional misrepresentation of data or information

CA Reports include the following components:

- Description of the problem - how it was identified and the date it was identified
- Root cause
- Description of the significance or consequences of the deviation– include sample ID number(s) affected
- CA(s) taken, including the timetable for implementation
- Actions implemented to prevent recurrence;
- Technicians/staff names (or job titles) involved
- Who prepared the report
- A review process with signatures and dates that includes a manager(s)

The TCEQ will review each CA report to determine if actions taken to resolve the deviation are acceptable. If CAs taken by a laboratory are unacceptable to the TCEQ, the TCEQ may not use sample results from the laboratory until such time that acceptable CA is achieved.

Corrected data must be submitted in a completely separate file from routinely submitted data. The laboratory must notify the TCEQ in advance in order to prevent duplication in the database of record.

Exhibit 1: Microbial Reporting Form (MRF 10525)

Exhibit 2: Microbial Monitoring Positive Result Report Form